

CAP-1642

#3

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant : Shuping Tong et al.
Serial No. : 09/818,066
Filed : March 27, 2001
Title : HEPADNAVIRUS PRE-S PROTEIN FRAGMENTS

Art Unit : 1642
Examiner : Unknown

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BOX SEQUENCE

Commissioner for Patents
Washington, D.C. 20231

RESPONSE TO NOTICE TO COMPLY WITH REQUIREMENTS
FOR PATENT APPLICATIONS CONTAINING
NUCLEOTIDE AND/OR AMINO ACID SEQUENCES

In response to the communication dated May 29, 2001 (copy enclosed), applicants submit herewith a Request to Under 37 CFR §1.821(e) to Transfer Computer-readable Form of the Sequence Listing for the instant application. In addition, applicants submit a paper copy of the Sequence Listing from the parent application.

Furthermore, applicants request entry of the following amendments.

In the specification:

Insert the paper copy of the Sequence Listing following the Oath/Declaration.

Replace the paragraph beginning at page 36, line 15, with the following rewritten paragraph:

--Cloning of p120 cDNA. Since amino acid sequences of all four p120 peptides perfectly matched chicken and human glycine decarboxylase gene, duck cDNA for p120 was isolated by a cDNA fragment of the chicken glycine decarboxylase. Chicken liver mRNA was reverse transcribed by random hexamers and superscript II reverse transcriptase (Gibco/BRL) at 42°C. The RNA template was removed by treatment with RNase H. The 1st strand cDNA was used as template for PCR amplification of the partial coding sequence for chicken glycine

CERTIFICATE OF MAILING BY FIRST CLASS MAIL

I hereby certify under 37 CFR §1.8(a) that this correspondence is being deposited with the United States Postal Service as first class mail with sufficient postage on the date indicated below and is addressed to the Commissioner for Patents, Washington, D.C. 20231.

July 30, 2001

Date of Deposit

Signature

Jennifer H. Payne

Typed or Printed Name of Person Signing Certificate

09/818,066

decarboxylase. The primers used were based on published sequence (Kume et al., 1991) and had the sequence 5'-ATCACTGAGCTCAAATTACCCCATGAGATG-3' (SEQ ID NO:14; sense primer, positions 679-701) and 5'-GGAAACTCGAGCTGGAAGCAGTGTTATGAA-3' (SEQ ID NO:15; antisense primer, positions 3038-3009). After 35 cycles of amplification using Vent DNA polymerase the product (2.3 kb) was purified from the agarose gel and labeled with ^{32}P -dCTP using random DNA labeling kit (Amersham). To screen for p120 cDNAs, oligo dT primed duck liver library was plated onto NZY plates at a density of 5×10^4 pfu/150 mm plate and cultured at 37°C overnight. The plaques were transferred onto duplicate nitrocellulose filters. The filters were hybridized with the ^{32}P labeled chicken cDNA fragment, washed and exposed to X-ray films. The final wash consisted of 0.5 x SSC/0.1% SDS at 55°C. For secondary screening, areas of NZY agar plates corresponding to positive hybridization signals were cut out and immersed in SM buffer to elute the phages. The phages were used to infect XL1 Blue MRF⁺ cells and plated onto NZY plates at low density to prevent individual plaques from merging with each other. The duplicate filters were rescreened with the chicken cDNA probe and positive plaques identified and isolated.--

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Attorney's Docket No.: 00786-287004 / MGH-0960.3

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REMARKS

Applicants hereby submit that the enclosures fulfill the requirements under 37 C.F.R. §1.821-1.825. The amendments in the specification merely insert the paper copy of the Sequence Listing and sequence identifiers in the specification. No new matter has been added.

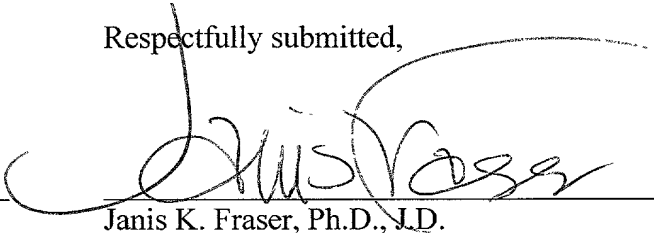
Attached hereto is a marked-up version of the changes made to the specification and claims by the current amendment.

Please apply any charges or credits to Deposit Account No. 06-1050.

Respectfully submitted,

Date:

July 30, 2001


Janis K. Fraser, Ph.D., J.D.
Reg. No. 34,819

Fish & Richardson P.C.
225 Franklin Street
Boston, MA 02110-2804
Telephone: (617) 542-5070
Facsimile: (617) 542-8906

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"Version With Markings to Show Changes Made"

In the specification:

Paragraph beginning at page 36, line 15, has been amended as follows:

Cloning of p120 cDNA. Since amino acid sequences of all four p120 peptides perfectly matched chicken and human glycine decarboxylase gene, duck cDNA for p120 was isolated by a cDNA fragment of the chicken glycine decarboxylase. Chicken liver mRNA was reverse transcribed by random hexamers and superscript II reverse transcriptase (Gibco/BRL) at 42°C. The RNA template was removed by treatment with RNase H. The 1st strand cDNA was used as template for PCR amplification of the partial coding sequence for chicken glycine decarboxylase. The primers used were based on published sequence (Kume et al., 1991) and had the sequence 5'-ATCACTGAGCTCAAATTACCCCATGAGATG-3' (SEQ ID NO:14; sense primer, positions 679-701) and 5'-GGAAACTCGAGCTGGAAGCAGTGTTATGAA-3' (SEQ ID NO:[22] 15; antisense primer, positions 3038-3009). After 35 cycles of amplification using Vent DNA polymerase the product (2.3 kb) was purified from the agarose gel and labeled with ³²P-dCTP using random DNA labeling kit (Amersham). To screen for p120 cDNAs, oligo dT primed duck liver library was plated onto NZY plates at a density of 5x10⁴ pfu/150 mm plate and cultured at 37°C overnight. The plaques were transferred onto duplicate nitrocellulose filters. The filters were hybridized with the ³²P labeled chicken cDNA fragment, washed and exposed to X-ray films. The final wash consisted of 0.5 x SSC/0.1% SDS at 55°C. For secondary screening, areas of NZY agar plates corresponding to positive hybridization signals were cut out and immersed in SM buffer to elute the phages. The phages were used to infect XL1 Blue MRF' cells and plated onto NZY plates at low density to prevent individual plaques from merging with each other. The duplicate filters were rescreened with the chicken cDNA probe and positive plaques identified and isolated.

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